The effects of single and repeat bleaching on photosynthesis, respiration, and feeding in three species of Caribbean coral

A senior Honors Thesis

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Abstract

Bleaching events are predicted to increase in frequency and intensity as a result of rising sea surface temperatures. Paired fragments of the Caribbean corals *Montastraea faveolata, Porites astreoides*, and *Porites divaricata* were experimentally bleached (treatment) or nonbleached (control) in outdoor flow-through seawater tanks. Half of the fragments were immediately collected, and half were returned to the reef to recover for one year at ambient temperature, followed by repeat bleaching the following summer. Our findings show that the mounding coral *P. astreoides* is the most tolerant, and the branching coral *P. divaricata* is the least tolerant, of single bleaching. Unexpectedly, it is the branching *P. divaricata* that appears to be the most tolerant of repeat bleaching and indicates that the underlying mechanisms for bleaching resilience are fundamentally different in repeat bleached corals compared to singly bleached corals. This study provides insight into how coral species' diversity and abundance could shift on Caribbean coral reefs in the coming decades.

Introduction

Coral reefs are declining globally due to a combination of direct and indirect human impacts (Hughes et al. 2003); (Veron et al. 2009). Mass coral bleaching events, a phenomenon where whole communities of corals loose a significant proportion of their vital endosymbiotic algae (commonly called zooxanthellae) and/or their algal photosynthetic pigments, are largely caused by elevated sea-surface temperatures (Jokiel and Coles 1990); (Glynn 1996); (Brown 1997); (Hoegh-Guldberg 1999); (D'Croz et al. 2001). The translocation of photosynthetically fixed carbon from these zooxanthellae (*Symbiodinium* spp.) is crucial to the energy budgets of most reef corals (Muscatine 1990). This mutualism has been the key to the evolutionary and ecological success of reef-building corals since the Triassic (Stanley 2003). While these symbiotic cnidarians

have shown resiliency through geological time, recent anomalies in global seawater temperature reveal their vulnerability to environmental stressors. The long-term impacts of bleaching include decreased growth in coral tissue and skeletal formation, reduction or cessation of gametogenesis and fertilization, and increased susceptibility to disease (Szmant and Gassman 1990); (Fitt et al. 1993); (Ward et al. 2000); (Omori et al. 1999). Extended and/or more extreme warming episodes can lead to mass coral mortality and ecosystem degradation (Wilkinson 2000; Stanley 2003). At the current rate of predicted global warming, mass bleaching events are expected to increase in frequency and severity in all tropical oceans in the coming decades, resulting in up to 60% coral mortality (Hoegh-Guldberg 1999; Wilkinson 2000; Buddemeier et al. 2004; Wooldridge et al. 2005). The Caribbean is expected to be especially affected as it appears to be extremely sensitive to seawater temperature increases of less than +1^{o}C (McWilliams et al. 2005), and is predicted to suffer bleaching events biannually within the next 20-30 years (Donner et al. 2007).

While our understanding of the effects of single bleaching grows, how corals respond to repeated bleaching remains unknown. Responses of coral species to single bleaching events may not be good predictors of the responses to repeated annual bleaching. The physiological impact on corals and the ecological stability of reefs could be dramatically different following repeat bleaching in ways that results from single bleaching studies do not reveal.

Corals can have dramatically different responses to elevated temperature stress: some corals may bleach and die, others may bleach and recover, and some do not visibly bleach at all (e.g., (Fisk and Done 1985; Oliver 1985; Ghiold and Smith 1990; Edmunds 1994; Marshall and Baird 2000; Stimson et al. 2002; Grottoli et al. 2004a). The underlying causes of variation in bleaching susceptibility and recovery is associated

with several factors including zooxanthellae density (Stimson et al. 2002), coral morphology (e.g., mounding versus branching) (Gleason 1993; Marshall and Baird 2000; Loya et al. 2001), energy reserves management (Grottoli et al. 2004a; Grottoli et al. 2006; Rodrigues and Grottoli 2006a,b; Anthony et al. 2009), and the ability to dramatically increase feeding rates and restore energy reserves when bleached (Grottoli et al. 2006; Palardy et al. 2008). Most importantly, much of this research has been conducted on only on a limited number of species, and the concept of trophic plasticity in response to bleaching (i.e., switching reliance from autotrophy to heterotrophy) has only been tested on three species of Pacific corals. Here, the effect of single and repeat bleaching on photosynthesis, respiration, and feeding rates of three species of Caribbean corals are examined. In the Caribbean, where increasing global seawater temperatures are coupled with the rapid decline of coral reefs (Gardner et al. 2003), the need to investigate the potential for recovery from bleaching and repeat bleaching are critical to determining the long-term resilience of these reefs.

Methods

Experimental Design – Single Bleaching

On 18 June 2010, two fragments from nine parent colonies of three Caribbean coral species—the mounding species *Montastraea faveolata* and *Porites astreoides*, and the branching species *Porites divaricata* – were collected from the reefs near Puerto Morelos, Mexico at depths ranging from 3-5 m (Table 1). Each fragment was mounted on three-inch diameter hexagonal tiles, and allowed to acclimate for 10 days (Fig. 1). The fragments were placed in outdoor aquarium tanks provided with blue-floss filtered flow-through seawater and allowed to acclimate for 10 days. The tanks were shaded with neutral density mesh to reflect natural light levels at the collection depth. One

fragment from each colony was assigned to the elevated temperature treatment (single bleaching treatment) and one was assigned to the ambient temperature control (control). On 28 June 2010, the temperature in the treatment tanks was gradually increased to 31.18 ^{o}C ± 0.01 ^{o}C (single bleaching treatment) over the course of 3 days, while the temperature in the control tanks remained unchanged with an average of 29.46 ^{o}C ± 0.01^{o}C. After 19 days, the temperature in the single bleached tanks was gradually reduced to ambient levels over the course of 2 days. Photosynthesis (P), respiration (R), and feeding rates were then measured on each coral fragment according to the methods described below.

Experimental Design – Repeat Bleaching

In July 2009, two fragments from nine parent colonies of the same three Caribbean coral species as above were collected from the reefs near Puerto Morelos, Mexico at depths ranging from 2.5-8 m (Table 1). Each fragment was mounted and placed in shaded outdoor tanks as above (Fig. 1). The seawater in half of the tanks was gradually increased to an average temperature of 31.5 ^{o}C ± 0.006 ^{o}C over the course of 3 days, while the temperature in the other half of the tanks remained ambient (30.7 ^{o}C ± 0.004 ^{o}C). After 15 days at the experimental temperatures, all of the fragments were returned to the reef to recover for one year at ambient temperature.

One year later on 18 June 2010, all of the fragments were retrieved from the reef, placed back in the same tanks, and allowed to acclimate for 10 days. Elevated temperature treatment fragments from 2009 were assigned to the elevated temperature treatment tanks in 2010 (repeat bleaching treatment). Ambient control fragments from 2009 were assigned to the ambient control tanks in 2010 (control). The temperature in repeat bleaching treatment tanks was gradually increased to an average of 31.6 ^{o}C ± 0.01 ^{o}C over the course of 3 days, while the temperature in ambient control tanks remained

at an average of 30.4 ^{o}C ± 0.01 ^{o}C After 18 days, the temperature in the repeat bleached tank treatments tanks was gradually reduced to ambient levels over the course of 2 days. Photosynthesis (P), respiration (R), and feeding rates were then measured on each coral fragment according to the methods described below.

Metabolic Rates

During the last two days of the single and repeat bleaching experiments, fragments were individually placed in Plexiglas chambers filled with seawater, the chambers were sealed shut, and all gaseous oxygen was purged. Chambers were placed in a Plexiglas tank filled with freshwater, which was temperature controlled using a heater/chiller and set to 30C°or 32°C for control and treatment fragments, respectively. Photosynthesis (P) and respiration (R) rates were determined from the change in [O_2] in the chambers using D901 Miniature Galvanic DO2 Probes from Qubit Systems. The probes were connected to a laptop, which ran LoggerPro software to record and analyze data. LoggerPro plotted dissolved oxygen concentration over time throughout each of the runs. P and R rates were determined by taking the slope of the LoggerPro graphs. The P and R rates were allowed to stabilize for the first 1-5 minutes of each run and only data collected after this stabilization period was used to calculate the P and R rates.

For P and R runs during the day, respiration was determined by covering the chambers with black plastic and allowing for the fragments to establish a constant respiration rate (usually 10 minutes). Shading was removed and an array of LED lights was turned on exposing the corals to 415µmol quanta $m^{-2}s^{-1}$ of light to induce maximal photosynthesis (usually 10-15 minutes). After a photosynthetic rate was established, corals were removed from the chamber and placed in their respective tanks. At night (approximately 8:00 PM), evening respiration rates were determined for the same corals. The same procedure as above was carried out, but only for respiration.

Feeding Rates

Coral fragments were placed on the reef in the morning and covered by a chamber for 8 hours during the day. The chamber covered all corals and was constructed of plastic and 50 micron mesh to allow flow, but prevent zooplankton from entering (Palardy et al. 2005). At night, after 8 hours of starvation during the day, the chambers were removed for 1 hour to allow the corals to feed. After one hour the corals were immediately collected and fixed in formalin to prevent further digestion of zooplankton. Within 48 hours a number of polyps on each fragment were dissected (as per (Palardy et al. 2005) (for *M. faveolata* and *P. astreoides* 150 polyps, or all polyps if the fragment had less than 150 were dissected. For *P. divaricata* all polyps were dissected as the polyps were relatively large and shallow) and number of zooplankton eaten per polyp and prey types were determined. Feeding rates were standardized to plankton captured/hour/cm^2 using the foil technique (Marsh 1970). Total number of polyps per fragment was calculated based on the number of polyps per cm^2 and the total surface area of the fragment. Feeding rate per number of polyps dissected was scaled up to feeding rate per fragment by using a scalar calculated by dividing the total number of polyps over the number of polyps dissected. The feeding rate per number of polyps counted was multiplied by this scalar to determine the feeding rate per fragment. The feeding rate for the whole fragment was then standardized to tissue biomass of each coral.

All measurements were made on whole coral samples (skeleton+ animal tissue+ zooxanthellae) ground with a mortar and pestle and normalized to total ash-free dry weight (tissue biomass of the organic fraction) according to Grottoli et al. (2004b).

CZAR

For each coral fragment, the total daily grams of photosynthetically fixed carbon per gram ash free dry weight (P_c) was calculated as the sum of net photosynthetically fixed carbon plus respired carbon during the day assuming a mole-to-mole relationship of CO_2 consumed (produced) to O_2 produced (consumed) during photosynthesis (respiration). Total daily respiration (R_c) was calculated as the sum of 12 hours of R_{day} plus 12 hours of R_{night}, CZAR (percent contribution of zooxanthellae-acquired carbon to daily animal respiration) for each coral was calculated as:

$$CZAR = \frac{Pc}{Rc} X100$$

CZAR is thus the percentage of a coral's daily metabolic energy demand that can be met through photosynthesis alone (Grottoli et al. 2006).

Statistical Analysis

Analysis of variance (ANOVA) compared the effects of species, genotype, temperature, and number of times bleached (single or repeat) on net P, day R, night R, CZAR, and feeding rates (Table 2). A posteriori slice tests (i.e., tests of simple effects, Winer 1971) determined if treatment and control averages significantly differed within species and between single and repeat bleachings. Bonferroni corrections were not used. Statistical analyses were generated using SAS software, Version 8.02 of the SAS System for Windows. (Copyright 1999–2001 SAS Institute Inc. SAS and all other SAS Institute Inc. product or service names are registered trademarks or trademarks of SAS Institute Inc., Cary, NC.) Values of $p \leq 0.05$ were considered significant.

Results

After 2.5 weeks at increased temperature (single bleaching), 88.9% of *M. faveolata* showed visible signs of bleaching (pale, or white) relative to controls, with 33.3% being completely bleached (Table 3). 33.3% of treatment *P. astreoides* and *P. divaricata* showed visible signs of bleaching relative to controls, with 33.3% and 11.1% being completely bleached, respectively.

After repeat bleaching (a second round of 2.5 weeks at increased temperatures), only 44.4% of *M. faveolata* showed visible signs of bleaching relative to controls. Interestingly 100% of *P. astreoides* and 57.1% of *P. divaricata* showed visible signs of bleaching. No fragments were fully bleached.

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Net Photosynthesis

Net P was significantly lower in single and repeat bleached *M. faveolata* and single bleached *P. divaricata* relative to controls (Fig. 2A, C) (Table 2). Net P was not significantly lower in single or repeat bleached *P. astreoides* relative to their controls (Fig. 2B).

Respiration

Day respiration (R_{day}) did not differ significantly between treatment and control *M. faveolata* fragments after single or repeat bleaching (Fig. 2D) (Table 2). In *P. astreoides* and *P. divaricata,* R_{day} was significantly lower in treatment corals compared to controls after single bleaching, but not after repeat bleaching (Fig. 2E, F) (Table 2). Night respiration (R_{night}) did not differ significantly between treatment and control fragments in any species after single, or repeat bleaching (Fig. 2G-I) (Table 2).

Feeding Rates

Feeding rates did not significantly differ between treatment and control coral fragments after single or repeat bleaching in both *M. faveolata* and *P. divaricata* (Fig. 2M, O) *(*Table 2*)*. However, feeding rates increased significantly in single bleached treatment *P. astreoides* compared to controls, but did not differ significantly after repeat bleaching (Fig. 2N) (Table 2).

CZAR

CZAR decreased significantly in treatment *M. faveolata* compared to controls after both single and repeat bleaching (Fig. 2J) (Table 2). CZAR did not significantly differ between treatment and control *P. astreoides* after single or repeat bleaching (Fig. 2K) (Table 2). In *P. divaricata*, CZAR significantly decreased in treatment coral fragments compared to controls after single bleaching, but not after repeat bleaching (Fig. 2L) (Table 2).

Discussion

P and R (metabolism)

The significant decreases in net P observed in singly bleached *M. faveolata* and *P. divaricata* were not unexpected as both species showed visible signs of bleaching after single bleaching (Table 3). Decreases in net P in bleached corals have been consistently observed in other studies (i.e., Porter et al. 1989, Grottoli et al. 2004, 2006, 2007). However, the lack of any change in net P in bleached *P. astreoides* was surprising since it had started to expel algal endosymbionts and had become visibly pale by the end of the single bleaching. Variation in net P rates in bleached coral species has also been reported in Hawaiian corals (Rodrigues & Grottoli 2007; Palardy et al 2008), Caribbean corals (Rodriguez-Román et al. 2006) and Pacific corals (Loya et al. 2001; Hoegh-

Guldberg 2004). When repeat bleached, coral net P significantly decreased in *M. faveolata*, but not in *P. astreoides* or *P. divaricata* (Fig. 2A-C). Forthcoming analyses on the algal symbiont density and chlorophyll *a* concentration will shed light on whether algal symbionts were present in surplus in the previously bleached corals as has been suggested by others (Dubinsky and Achituv 1990; Stambler and Dubinsky 2005; Rodrigues and Grottoli 2007).

Though R_{night} was not affected by single or repeat bleaching (Fig. 2G-I), the significant decrease in R_{day} in singly bleached *P. astreoides* and *P. divaricata* corals (Fig. 2D-F) indicates a decrease in metabolic activity for these species (Rodrigues and Grottoli 2007). However, none of the species reduced their metabolic rates when repeat bleached (Fig. 2D-F). This is not surprising for *P. astreoides* and *P. divaricata* corals as their ability to acquire photosynthetically fixed carbon was unaffected by repeat bleaching (Fig. 2E, F). However, for *M. faveolata*, there was a significant decrease in photosynthetically fixed carbon after both single and repeat bleaching, but there was no compensation for this loss of carbon by reducing metabolic rate. In order to survive, bleached *M. faveolata* must utilize another strategy to compensate for a decrease in photosynthetically acquired carbon, such as catabolizing energy reserves or increase heterotrophy (Grottoli et al. 2006; Rodrigues and Grottoli 2007).

CZAR

In agreement with previous publications, all three species had CZAR values >100% when non-bleached (controls) indicating that all three species met 100% of their daily metabolic demands photoautotrophically when healthy (Fig. 2J-L) (Muscatine et al. 1981; Grottoli et al. 2006). When singly bleached, none could meet 100% of their daily metabolic demand via photosynthesis alone (Fig. 2J-L), and other strategies must

be utilized. These results are in agreement with data from Hawaiian corals subjected to single bleaching (Grottoli et al. 2006).

Feeding

Feeding rates did not significantly differ between singly bleached and control *M. faveolata* or *P. divaricata* (Fig. 2M, O), indicating that these two species did not increase heterotrophy to cope with the reduction in photosynthetically acquired carbon (Grottoli et al. 2006); (Palardy et al. 2006). To survive, they would have had to catabolize energy reserves (Grottoli et al. 2006; Rodrigues and Grottoli 2007), or as in the case of *P. divaricata* reduce metabolic demand (Fig. 2F). Feeding rates significantly increased in singly bleached *P. astreoides* compared to controls, indicating that this species increased heterotrophy to help offset loses of carbon during bleaching (Grottoli et al. 2006; Palardy et al. 2006). In contrast, none of these three species increased feeding to compensate for loss of photosynthetic carbon after repeat bleaching. The need to increase feeding in *P. astreoides* and *P. divaricata* may not have been great since there was not a significant loss of photosynthetic carbon after repeat bleaching in these species (Fig. 2B, C), so there is no reason to compensate. However, there was not a significant loss of photosynthetic carbon in single bleached *P. astreoides*, yet feeding rates increased (Fig. 2B, N). As previously shown in Grottoli et al (2006), corals often fix far in excess of their daily metabolic carbon needs. For *M. faveolata*, CZAR and net P were significantly lower in repeat bleached corals than in controls, indicating that the daily metabolic needs of the corals were not being met by photosynthesis alone. Increasing feeding would help solve this problem, but since feeding rates did not increase the corals must turn to another energy source, such as energy reserves (Grottoli et al. 2006), (Rodrigues and Grottoli 2007). Energy reserves are finite, and once depleted, if the corals have not recovered their photosynthetic pathways, they will die.

This means that *M. faveolata* is likely the most at risk of these three species during repeat bleaching events.

Summary

P and CZAR decreased while R_{day}, R_{night} (Fig. 2 A, J) and feeding rates (Fig. 2 M) did not in single and repeat bleached *M. faveolata* corals. Thus metabolically, this species responds similarly to both single and repeat bleaching, is susceptible to both single and repeat bleaching, and is most at risk of the three species studied as the frequency and magnitude of bleaching events continue to increase as a result of rising sea surface temperatures.

P, R_{night}, and CZAR in *P. astreoides* corals were unaffected by single or repeat bleaching (Fig 2B, H, K, N). However, dramatic increases in feeding rates appear to have played a critical role in mediating the negative effects of single bleaching and rendered this species the most resilient to single bleaching. As feeding did not increase when repeat bleached, it appears that *P. astreoides* is less resilient to repeat bleaching than to single bleaching.

P, R_{day}, and CZAR decreased in singly bleached, but not in repeat bleached *P. divaricata* (Fig 2C, F, L). In the absence of any compensation in feeding rates (Fig 3O), this species appears to be susceptible to single bleaching, but resilient to repeat bleaching. Unexpectedly, the branching coral species was more resilient to repeat bleaching than the two mounding species. This is contrary to all published findings to date that tend to highlight the higher resilience of mounding corals compared to branching corals (Loya et al. 2001). Additional research is needed to determine if branching Pacific corals are also more tolerant of repeat bleaching than their mounding counterparts.

Future Work

In the future percent carbon of local zooplankton samples must be determined in order to calculate CHAR (percent contribution of heterotrophically acquired carbon to daily animal respiration). This, coupled with CZAR and feeding will help completely explain how the daily metabolic energy needs of each species are met (or not) after single and repeat bleaching events. Also, this data can be compiled with energy reserve, symbiont, and DOC data to explain how coral host and algal physiology changes in these three species after bleaching events, and how/why that response differs between single and repeat bleaching events.

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Tables and Figures:

Table 1: Parent colony collection dates, depths, and locations for single bleaching and repeat bleaching fragments.

Species	Genotype	Date	Depth	Location	Coordinates
Single Bleaching					
P. astreoides	1-9	6/18/10	3.048 m	El Islote	20°55.607'N, 86°49.882'W
P. divaricata	1-9	6/18/10	3.048 m	El Islote	20°55.607'N, 86°49.882'W
M. faveolata	5,9	6/18/10	4.876 m	The Wall	20°49.432'N, 86°52.664'W
M. faveolata	8	6/18/10	4.876 m	Jardines	20°50.045'N, 86°52.694'W
M. faveolata	1-4, 6, 7	6/18/10	4.572 m	Radio Pirata	20°51.260'N. 86°51.909'W
Repeat Bleaching					
P. astreoides	1-9	7/4/09	3.048 m	El Islote	20°55.607'N, 86°49.882'W
P. divaricata	1-9	7/5/09	2.743 m	El Islote	20°55.607'N, 86°49.882'W
M. faveolata	5,9	7/6/09	7.924 m	The Wall	20°49.432'N, 86°52.664'W
M. faveolata	8	7/9/09	3.962 m	Jardines	20°50.045'N, 86°52.694'W
M. faveolata	1-6, 6, 7	7/9/09	2.438 m	Radio Pirata	20°51.260'N. 86°51.909'W

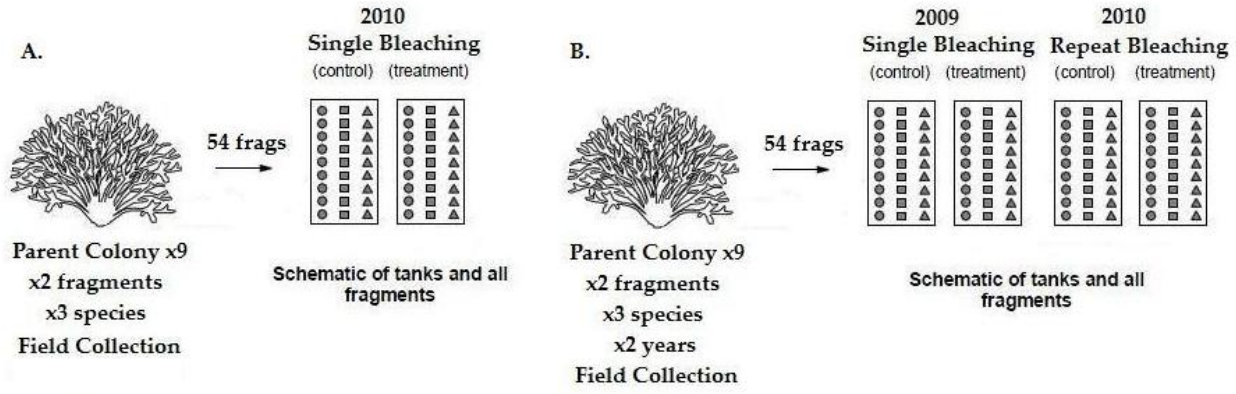

Figure 1: Schematic of the experimental design. **A:** in June 2010, nine newly collected fragments of each species were placed in control and treatment tanks for 2.5 weeks in the same manner as the previous fragments were**. B:** Nine fragments of each species: *M. faveolata* (●), *P. astreoides* (■), and *P. divaricata* (▲) were placed in control and treatment tanks (June 2009). After 2.5 weeks in the tanks the fragments were placed back on the reef to recover. In June 2010, the fragments were again experimentally bleached.

Table 2. Results of six three-way ANOVAs for average photosynthesis rate (F= 6.21, p< 0.0001), day respiration rate (F= 2.63, p= 0.0003), night respiration rate (F= 2.13, p= 0.0038), CZAR (F= 1.73, p= 0.0263), feeding rate (F= 1.45, p= 0.0944) comparing 9 colonies or genotypes, at two temperatures (ambient (30.3 °C and 31.3°C), after single or repeat bleaching within three species (M. faveolata, P. astreoides, and P. divaricata). Effects of temperature (T) were fixed and fully crossed. Genotype (G) was a random effect. Interaction terms involving genotype were combined with the residual. Yr, times bleached (1= single, 2= repeat), T, temperature. df, degrees of freedom; SS sum of squares of the effect.

	Effect	df	SS	F-statistic	p-value
Net P	Yr	1	0.24572979	5.76	0.0191
	T	1	2.42827812	56.93	<.0001
	sp	2	0.53014505	6.21	0.0033
	Geno (sp)	24	1.88639757	1.84	0.0257
	Yr*T	1	0.34325507	8.05	0.0060
	Yr*sp	2	2.74899734	32.22	<.0001
	T*sp	2	0.41650892	4.88	0.0104
	Yr*T*sp	2	0.60298084	7.07	0.0016
Day R	Yr	1	0.01569639	0.33	0.5668
	T	1	0.39498366	8.33	0.0052
	sp	2	0.44227574	4.67	0.0126
	Geno (sp)	24	1.56576692	1.38	0.1527
	Yr*T	1	0.10405168	2.20	0.1430
	Yr*sp	2	1.51682997	16.00	<.0001
	T*sp	2	0.07520225	0.79	0.4564
	Yr*T*sp	2	0.14516552	1.53	0.2235
Night R	Yr	1	0.01516970	0.37	0.5458
	T	1	0.01439617	0.35	0.5562
	sp	2	0.78834236	9.57	0.0002
	Geno (sp)	24	1.01634720	1.03	0.4451
	Yr*T	1	0.38292450	9.30	0.0032
	Yr*sp	2	0.64302580	7.81	0.0009
	T*sp	2	0.06408133	0.78	0.4632
	Yr*T*sp	2	0.01904962	0.23	0.7941
CZAR	Yr	1	14.96020	0.00	0.9449
	T	1	59893.57888	19.24	<.0001
	sp	2	7959.20361	1.28	0.2850
	Geno (sp)	24	78681.50868	1.05	0.4176
	Yr*T	1	2120.29929	0.68	0.4121
	Yr*sp	2	35822.17751	5.75	0.0049
	T*sp	2	10897.91491	1.75	0.1814
	Yr*T*sp	2	892.29857	0.14	0.8667
Feeding Rates	Yr	1	1225.93508	1.77	0.1876
	T	1	1117.23275	1.61	0.2081
	sp	2	6536.64412	4.72	0.0120
	Geno (sp)	24	16651.19754	1.00	0.4753
	Yr*T	1	564.51530	0.82	0.3696
	Yr*sp	2	4488.50373	3.24	0.0451
	T*sp	2	2368.25128	1.71	0.1882
	Yr*T*sp	2	1806.31463	1.31	0.2777

Table 3: Percent mortality for each species and treatment after single and repeat bleaching. nonbleached= brown and covered in living tissue, partially bleached= either 100% of the tissue was pale or yellow, or some of the tissue was bleached and some was healthy, bleached= 100% white or 50% white and the rest was pale/yellow, n=9, MF= *Montastraea faveolata*, PA= *Porites astreoides*, PD= *Porites divaricata*

Single Bleaching, June 2010					
		Nonbleached	Partially Bleached	Bleached	Dead
Summary:	MF control	100.0	0.0	0.0	0.0
%	MF treatment	11.1	55.6	33.3	0.0
	PA control	100.0	0.0	0.0	0.0
	PA treatment	66.7	0.0	33.3	0.0
	PD control	100.0	0.0	0.0	0.0
	PD treatment	66.7	22.2	11.1	0.0
Repeat Bleaching, June 2010					
		Nonbleached	Partially Bleached	Bleached	Dead
Summary:	MF control	100.0	0.0	0.0	0.0
%	MF treatment	44.4	44.4	0.0	11.1
	PA control	100.0	0.0	0.0	0.0
	PA treatment	0.0	100.0	0.0	0.0
	PD control	100.0	0.0	0.0	0.0
	PD treatment	42.9	57.1	0.0	0.0

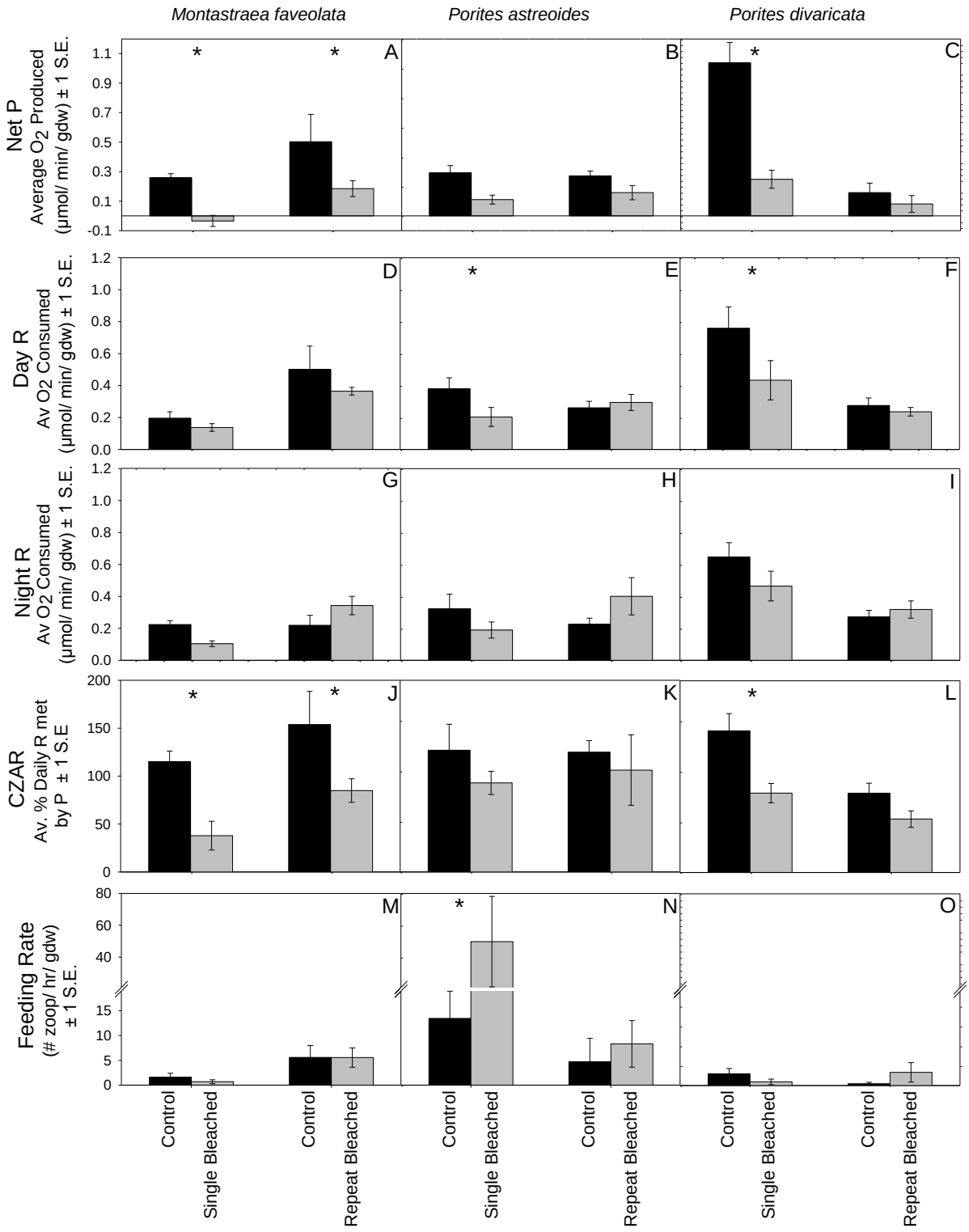

Figure 2: (A-C) Net photosynthesis (P), (D-F) day respiration (R), (G-I) night R, (J-L) CZAR (percent contribution of zooxanthellae acquired carbon to daily animal respiration), and (M-O) feeding rate for single and repeat bleached *Montastraea faveolata*, *Porites astreoides*, and *Porites divaricata*. zoop= zooplankton, hr= hour, S.E. = standard error, Av.=average, gdw = ash free gram dry weight